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ORIGINAL ARTICLE

Association of dietary calcium, phosphorus, and magnesium intake with caries status among schoolchildren

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Abstract The aim of this study was to investigate the associations between caries experience and daily intake of calcium (Ca), phosphorus (P), magnesium (Mg), and Ca/P ratio. A total of 2248 schoolchildren were recruited based on a population-based survey. Each participant received a dental examination and questionnaire interviews about the 24-hour dietary recalls and food frequency. The daily intake of Ca, P, Mg, and Ca/P ratio were inversely associated with primary caries index, but only the Ca/P ratio remained significant after adjusting for potential confounders. According to the Taiwanese Dietary Reference Intakes, the Ca/P ratio was related to both caries in primary teeth (odds ratio = 0.52, $p = 0.02$) and in permanent teeth (odds ratio = 0.59, $p = 0.02$). The daily intakes of Ca/P ratio remained an important factor for caries after considering potential confounding factors.

Conflicts of interest: All authors declare no conflicts of interest.

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Introduction

Nutritional factors are generally associated with several systemic diseases, gastrointestinal disorders, most cancers, as well as oral diseases [1]. The most common chronic oral disease, dental caries, may lead to pain from untreated teeth, reduced chewing ability, nutritional insufficiencies, lower self-esteem, and the problems of speaking and learning in schoolchildren [2].

Dental cavities are caused by the demineralization of tooth enamel by acids, such as lactate and acetate, formed from the fermentation of sugars and starches by dental plaque bacteria. *In vitro* studies show that milk or dairy products can reduce enamel solubility, promote its remineralization, and prevent the adhesion of mutans streptococci to the tooth surface [3]. In terms of the relationship between dietary minerals intake and oral disease, the calcium (Ca) and phosphorus (P) concentrations of dental plaque and the levels of Ca and P ions in the saliva could affect the balance between demineralization and remineralization of enamel [4]. Some epidemiological studies have revealed that humans with relatively high Ca and P in their plaque experience correspondingly lower caries [5]. In addition, Stanton [6] found that the dietary Ca/P ratio was strongly associated with caries development in human teeth, and indicated the effect of anticariogenicity when the dietary Ca/P ratio was 1.1 or 1.2. Stanton [6] also inferred that the dietary Ca/P ratio could influence the enzymic activity in oral bacteria. However, recent studies have disagreed and shown a less important role of the Ca/P ratio in caries development. Some epidemiological investigations have revealed there is no significant relationship between caries and dietary Ca/P ratio [7]. Similarly, magnesium (Mg) has also been shown to have both significant [8] and nonsignificant [9] associations with tooth decay.

The major food sources of minerals have been reported by Wu et al. [10] for Taiwanese schoolchildren. The dietary intake of Ca was 31.5% from dairy products (fresh milk, yogurt, cheese, and other dairy products) and 10.3% from dark-green/yellow vegetables. For the intake of P, dairy products and pork/pork products contributed about 14.9% and 12.0%, respectively. Mg intake was mainly from rice/rice products (10.5%).

An earlier study in Taiwan showed that daily milk consumption was significantly associated with a lower risk of permanent teeth decay in schoolchildren [11]. However, Yen et al. [12] indicated that neither dietary intake of Ca and P nor dietary Ca/P ratio was significantly associated with dental caries in preschool children. Therefore, the purpose of this research was to investigate the associations among caries experience and intakes of Ca, P, Mg, and Ca/P ratio in a population based cross-sectional study of elementary schoolchildren.

Methods

Study participants and study design

This study used the database of the Nutrition and Health Survey in Taiwan Elementary School Children 2001—2002 (NAHSIT Children 2001—2002), which is a population-based survey investigating the nutrition and health status of elementary schoolchildren in Taiwan. Towns and districts were classified into 13 strata by particular ethnic and geographical characteristics, which included Hakka areas, mountain areas, eastern Taiwan, the Penghu Islands, three northern regions, three central regions, and three southern regions. One hundred and four schools were selected (8 schools from each stratum), and within each school, 24 students were randomly selected (4 students from each grade) based on the "probabilities proportional to sizes" approach. The survey included face-to-face interviews and physical examinations (including dental examination). A total of 2248 schoolchildren (1196 boys and 1052 girls) aged 6—12 years participated in this survey and completed both interviews and dental examination. Informed consent was signed by one of the parents of each participating child. This study was approved by the Institutional Review Board (Human Experiment and Ethics Committee, Kaohsiung Medical University Hospital, MUH-IRB-990027). More detailed description of the sampling design is provided in Tu et al.'s report [13].

Dental examination

Each participant received a dental examination and a personal interview. We followed the guidelines of the *Oral Health Surveys—Basic Methods* (4^{th} edition) for dental examinations with dental mirrors and CPI (Community Periodental Index) probes. Three dentists participated in the survey. The presurvey calibration practice yielded 0.79, 0.80, and 0.90 for the kappa coefficients on caries diagnosis. During the survey, 5% of the students were reexamined by a second dentist, and the kappa coefficients were 0.77, 0.80, and 0.82. Student caries status was recorded as the DMFT index [D (decayed), M (missing due to caries), F (filled)] of the permanent teeth and the deft index [d (decayed), e (indicated for extraction), f (filled)] of the primary teeth. The DMFT and deft indexes were further categorized into three groups, in which Group 1 was for caries index being 0, Group 2 was for caries index being 1—3, and Group 3 was for caries index being 4 or higher.

Questionnaire

Questionnaire information, which included sociodemographics, tooth brushing habits, fluoride exposure, 24-hour dietary recalls, and food frequency, was collected by trained interviewers. Body mass index (BMI) was also obtained from the physical examination. Dietary mineral (Ca, P, and Mg) intake was assessed using data from 24-hour dietary recalls during household questionnaire interview. Interviewers used several tools, including food piece models, abstract food models, measuring cups, spoons, and electronic weight, to help the participants recall the amount of foods individuals consumed. The collected information of food models were first transformed into weights of foods consumed, and then the amounts were calculated using the Assessment of Chinese Dietary Intake system, developed by the Institute of Biomedical Sciences and the Institute of Information Science, Academia Sinica [14]. The major nutrient databases included "Nutrient composition Data Bank for Foods in Taiwan Area" and "Composition Data Bank for Foods in

Taiwan" [14]. The food sources were grouped into 12 major items and 47 minor items for further analysis. More detailed description of 24-hour dietary recalls and important food sources of minerals (Ca, P, and Mg) for children have been reported in Wu et al.'s report [10]. The dietary intake information of snack and beverage was collected through food frequency questionnaire and was jointly answered by the parents and the child based on the child's actual eating patterns in the month prior to the survey. Frequency categories ranged from "never," "1–2 days/week," "3–4 days/week," "5–6 days/week," to "every day," and a number marked scale from 0 to 7 using 0.5 as one unit was presented underneath the five response items.

Statistical analysis

All statistical analyses were weighted according to the area/age/gender distribution in 2001–2002 to represent the general elementary schoolchildren in Taiwan. The primary outcome variables, which comprised the daily intake levels of Ca, P, Mg, and Ca/P ratio from various food sources, were calculated while adjusting for the design effect of the complex sampling scheme [10]. Chi-square tests were used to compare the distributions of three different groups of caries index (deft or DMFT: 0, 1–3, and 4 or more) among gender, age groups, and education level of their parents. The mean daily intakes of Ca, P, Mg, and Ca/P ratio among caries groups were first compared by analysis of variance with Dunnett *post hoc* comparison (using the group of no caries as the control group). In addition, the adjusted means were also computed based on the analysis of covariance models with both deft caries group and DMFT caries group in the model along with adjusting variables of sex, age, birth order, parental education levels, BMI, sweet intake, tooth brushing habits, and fluoride exposure. The associations between daily intakes of Ca, P, Mg, and Ca/P ratio were evaluated using Pearson's correlation coefficients.

Multiple logistic regressions were adapted to compute the adjusted odds ratios (ORs) for developing caries (deft or DMFT index larger than 0). The analysis variables included daily Ca, P, and Mg intake levels and Ca/P ratio. Each variable was dichotomized into two groups according to the Taiwanese Dietary Reference Intakes (DRIs) in 2002 to indicate whether schoolchildren's dietary intake fulfilled the recommended levels [10]. The statistical adjustment in multiple logistic regressions included sex, age, birth order, parental education level, BMI, sweet intake, tooth brushing habits, and fluoride exposure. Statistical analyses were conducted using SAS version 9.1 (SAS Institute Inc., Cary, NC, USA).

Results

There were 2248 schoolchildren included in this survey. The original number of participants and weighted prevalence rates of groups in two different caries indexes among gender, age, and education level of parents groups are shown in Table 1. There were significant differences in DMFT index groups between gender (proportions of DMFT = 0, 1–3, and 4+ were 52.3%, 27.5%, and 20.2%, respectively, in boys and 40.0%, 31.2%, and 28.8%, respectively, in girls; $p < 0.0001$). The distributions of both DMFT index groups and deft index groups were also significantly different ($p < 0.001$) among age groups (6–12 years old). Schoolchildren whose fathers had an education level of junior high school or lower had 42.4% with DMFT index = 0, 29.3% with DMFT index = 1–3, and 28.3% with DMFT index = 4. Correspondingly, 47.5%, 30.2%, and 22.3% had fathers with senior high school education, and 51.6%, 27.9%, and 20.5% had fathers with college and above education. These differences reached statistical significance in the Chi-square test ($p = 0.001$), and a similar trend was noted for mothers' education levels ($p = 0.001$).

To investigate the means of daily Ca, P, and Mg intake and Ca/P ratio among the different caries index groups (Table 2), the average daily Ca intake was decreased with deft index groups (from 560.75 to 496.52, $p = 0.002$), and the average daily P or Mg daily intake levels were also significantly decreased (P from 1135.96 to 1035.05, $p < 0.001$; Mg from 251.31 to 229.17, $p = 0.001$). In the DMFT index groups, only the Ca/P ratio was significantly associated with the lower value of Ca/P ratios. After adjusting for the confounding effects of sex, age, birth rank, parental education level, BMI, sweet intake, tooth brushing habits, and fluoride exposure, it was noted that the adjusted means from the analysis of covariance models was no longer significantly associated with DMFT/deft index groups except for the Ca/P ratio ($p = 0.02$ in the deft index group and $p = 0.04$ in the DMFT index group). Pearson correlation coefficients among variables showed that the dietary Ca, P, and Mg daily intake were significantly associated with each other except for P versus Ca/P (not shown in Table 2).

To investigate the effect of dietary intake variables on developing caries (deft or DMFT index above 0), logistic regressions were adapted. Each dietary intake variable was dichotomized into two groups according to the Taiwanese DRIs in 2002 to indicate whether the schoolchildren's dietary intake fulfilled the recommended levels [10]. In Table 3, each intake variable was first analyzed separately (univariate analysis), and then they were joined in the same models (multivariable analysis). All of the logistic regressions included sex, age, birth order, parental education level, BMI, sweet intake, tooth brushing habits, and fluoride exposure as part of adjusting variables in the analysis models.

When analyzing dietary intake variables in separate models, schoolchildren with sufficient Ca intake (above DRIs level) were significantly associated with a lower risk of developing caries in permanent teeth (DMFT > 0) [OR = 0.76, 95% confidence interval (CI) = 0.58–0.99, $p = 0.04$]. The Ca/P ratio showed a significantly lower risk of deft index above 0 (OR = 0.49, 95% CI = 0.30–0.78, $p = 0.003$), and the DMFT index above 0 (OR = 0.55, 95% CI = 0.37–0.82, $p = 0.004$). When analyzing Ca, P, Mg, and Ca/P ratio together (multivariable), only the daily intake of Ca/P ratio was significantly associated with a lower risk of developing caries in primary teeth (OR = 0.52, 95% CI = 0.30–0.90, $p = 0.02$) and in permanent teeth (OR = 0.59, 95% CI = 0.37–0.93, $p = 0.02$; Table 3).

Discussion

Only a few studies have assessed the influence of Ca, P, and Mg daily intake on dental caries of the mixed dentition in

Table 1 Prevalence of caries status in different sex, age and education level of parents groups.[a]

	Actual sample size	deft index of primary teeth				DMFT index of permanent teeth			
		0 teeth	1–3 teeth	4+ teeth	*p*	0 teeth	1–3 teeth	4+ teeth	*p*
	n	%	%	%		*n*	%	%	
Total sample size	2248	33.4	19.9	46.7		46.4	29.3	24.3	
Sex									
Boys	1196	32.9	20.3	46.8	0.83	52.3	27.5	20.2	<0.001
Girls	1052	33.9	19.5	46.6		40.0	31.2	28.8	
Age (y)									
6	187	13.1	14.5	72.4	<0.001	75.1	20.3	4.6	<0.001
7	388	14.0	12.6	73.4		68.8	21.0	10.2	
8	373	10.7	16.1	73.2		57.6	24.7	17.6	
9	360	15.2	28.2	56.6		40.4	36.8	22.8	
10	381	37.8	28.9	33.3		29.1	41.6	29.3	
11	380	71.0	22.0	7.0		30.8	29.3	39.9	
12	179	87.1	10.5	2.4		26.3	26.1	47.6	
Education level of father									
Junior high school and below	954	34.2	17.9	47.9	0.29	42.4	29.3	28.3	0.001
Senior high school	772	31.8	21.6	46.6		47.5	30.2	22.3	
College and above	522	34.3	20.9	44.9		51.6	27.9	20.5	
Education level of mother									
Junior high school and below	1076	32.9	19.3	47.8	0.55	41.9	32.2	25.9	0.001
Senior high school	838	34.1	21.3	44.6		48.9	27.1	24.1	
College and above	334	33.2	18.4	48.5		53.0	26.4	20.6	

deft = d (decayed), e (indicated for extraction), f (filled) of primary teeth; DMFT = D (decayed), M (missing due to caries), F (filled) of permanent teeth.

[a] Numbers were weighted according to the area/age/sex distribution in 2001–2002 Taiwan elementary schoolchildren population.

population-based studies. In general, 24-hour dietary recall is the most widely used method for obtaining valid quantitative recall data. It can provide detailed information on dietary intake and is suitable in face-to-face, computer-assisted, and telephone interviews [15]. However, because of its complexity, it is rarely used in general dental health-related population-based studies. The NAHSIT Children 2001–2002 was sponsored by the Department of Health in Taiwan. It was conducted to monitor the relationships between the health and dietary nutrition intake of schoolchildren. Hence, in the present study, we were able to use the 24-hour dietary recall information, which was measured by a professional nutrition team with sufficient sample size, from the database of NAHSIT Children 2001–2002 to analyze the association of daily mineral intake with caries status in schoolchildren.

Diet is one of the important factors in dental caries. Previous studies have indicated that daily consumption of such products as fresh milk is related to a lower risk of caries. In Taiwan, Yang et al. [11] found that daily milk consumption was significantly associated with a lower risk of permanent teeth decay in schoolchildren. One Italian study suggested that in 6–11-year-old schoolchildren, milk consumption was significantly correlated to lower caries experience [16]. In another cross-sectional survey, milk intake was shown to exhibit protective associations with reported-caries experience [17].

Milk contains anticariogenic substances, such as Ca, P, casein, proteins, and fats. The Ca concentration of dental plaque and the level of Ca and P ions in the saliva could affect the balance between demineralization and remineralization of enamel [18]. In Schamschula et al.'s [19] study, it was shown that the concentrations of Ca, P, Mg, lithium, strontium, and fluoride in plaque are inversely associated with the DMFT index [19]. Furthermore, based on Kashket and DePaola's [20] review, increase of Ca and P (i.e., from milk or cheese) should lead to increased remineralization; however, remineralization occurs naturally in the mouth when saliva contains Ca and P ions that are supersaturated with respect to dental enamel. Ca and P have also been shown to facilitate remineralization in early carious lesions, which therefore increases the resistance to caries development [21]. Furthermore, in Herod's [22] investigation, it seemed that high Ca and P content could be another factor in the anticariogenic mechanism of cheese. However, previous studies among children revealed that tooth decay was not observed to be correlated with the intake of Ca and P [8,23]. In the present study, a higher value of the deft index was associated with lower amounts of daily Ca or P intake. Because the associations were no longer significant after adjusting for potential confounding factors, one could speculate that the daily dietary intakes of Ca or P might not be the primary nutritional factors for caries.

In terms of Mg, Jawed et al. [9] indicated there was no significant difference in salivary Mg in participants as compared to controls, but another investigation [23] showed that there was a negative association between Mg

Table 2 Average daily intake of nutrients among DMFT/deft index groups.[a]

	Actual sample size (n)	Ca (mg/d)	P (mg/d)	Mg (mg/d)	Ca/P ratio
Original average					
deft index					
0 teeth	749	560.75 ± 441.39	1135.96 ± 587.68	251.31 ± 134.27	0.493 ± 0.249
1–3 teeth	433	524.70 ± 395.88	1075.12 ± 521.20	234.22 ± 132.80	0.478 ± 0.235
4+ teeth	1066	496.52 ± 347.12	1035.05 ± 486.79	229.17 ± 124.64	0.475 ± 0.213
ANOVA p		0.002	<0.001	<0.001	0.23
Dunnett *post hoc* comparison			0 vs. 4+	0 vs. 4+	0 vs. 4+
DMFT index					
0 teeth	977	537.74 ± 409.71	1060.36 ± 532.22	231.27 ± 126.14	0.499 ± 0.244
1–3 teeth	684	510.86 ± 381.68	1098.91 ± 552.47	245.85 ± 139.53	0.465 ± 0.212
4+ teeth	587	511.90 ± 369.21	1081.23 ± 501.00	239.61 ± 123.64	0.469 ± 0.223
ANOVA p		0.27	0.33	0.07	0.003
Dunnett *post hoc* comparison					0 vs. 1–3 0 vs. 4+
Adjusted means (mean ± SE)					
deft index					
0 teeth	749	567.35 ± 21.95	1102.14 ± 29.92	242.17 ± 7.34	0.521 ± 0.013
1–3 teeth	433	535.30 ± 23.52	1073.54 ± 32.05	233.57 ± 7.86	0.492 ± 0.014
4+ teeth	1066	519.85 ± 19.89	1077.04 ± 27.11	235.95 ± 6.65	0.481 ± 0.012
ANOVA p		0.13	0.64	0.55	0.02
Dunnett *post hoc* comparison					0 vs. 4+
DMFT index					
0 teeth	977	557.73 ± 19.09	1075.15 ± 26.01	233.37 ± 6.38	0.515 ± 0.011
1–3 teeth	684	535.15 ± 21.50	1105.75 ± 29.30	244.21 ± 7.18	0.488 ± 0.013
4+ teeth	587	529.62 ± 22.44	1071.81 ± 30.58	234.11 ± 7.50	0.490 ± 0.013
ANOVA p value		0.36	0.43	0.21	0.04
Dunnett *post hoc* comparison					0 versus 1–3

Data are expressed as mean ± SD unless otherwise indicated.
ANCOVA = analysis of covariance; ANOVA = analysis of variance; BMI = body mass index; Ca = calcium; deft = d (decayed), e (indicated for extraction), f (filled) of primary teeth; DMFT = D (decayed), M (missing due to caries), F (filled) of permanent teeth. Mg = magnesium; P = phosphorus.
[a] Adjusted means were computed from ANCOVA models with adjustment of variables (sex, age, birth order, parental education level, BMI, sweet intake, tooth brushing habits, and fluoride exposure).

intake and dental caries. In this study, similar to the results of Ca and P, the daily Mg intake only appeared significantly associated with the deft index prior to any adjustment of the confounding factors.

Considering the Ca/P ratio correlated with microhardness values in active lesions [24], the chronic consumption of a low Ca/P ratio diet would increase the secretion of parathyroid hormone, induce a persistently high parathyroid hormone (even within the normal range), and finally could reduce bone mass and density [25]. Some studies that have been conducted regarding dietary Ca/P ratio have shown conflicting results. Rugg-Gunn et al. [7] reported no correlations between tooth decay, dietary Ca and P intake, and Ca/P ratio in 405 students aged 11.5–13.5 years. Similar results were shown by Marques and Messer [23] and by Yen et al. [12]; however, Stanton [6] revealed that the variation in dietary Ca/P ratio was related to cariogenicity (particularly low P content) in 183 adult patients.

Our study results indicated that the Ca/P ratio was a significant factor for developing caries in both primary and permanent teeth. When analyzed together with known risk factors (sex, age groups, birth rank, parental education levels, BMI, sweet intake, tooth brushing habits, and fluoride exposure) and with daily intake of Ca, P, and Mg, the ratio of Ca/P remained as an independent factor for caries. The dietary Ca/P ratio seemed to be more important than Ca and P separately in dental caries of schoolchildren.

The NAHSIT Children 2001–2002 used a cross-sectional design. Therefore, we could not interpret the cause–effect relationship between minerals and tooth decay by the results from this analysis. Our findings only provided the relationships between current dietary behaviors and dental caries experience. Although the amounts of Ca, P, and Mg were shown to be associated with caries in several *in vitro* or *in vivo* studies, our results from a population-based study do not support that individual daily mineral intake can be considered a primary factor for caries development. In addition, a literature review indicated that concentrations of Ca, P, Mg, lithium, strontium, and fluoride in plaque are associated with caries [19]; however, it should be noted that this study only analyzed Ca, P, and Mg.

Ca and P are associated with caries experience in terms of the ratio of Ca/P. Given that the dairy products are the major food sources both for Ca (31.5%) and P (14.9%), the

Table 3 Odds ratio of caries by DRIs of daily Ca, P, and Mg and Ca/P intake in Taiwanese elementary school children.[a]

	Univariate		Multivariate	
	1+ teeth versus 0 teeth		1+ teeth versus 0 teeth	
	OR (95% CI)	*p*	OR (95% CI)	*p*
deft index of primary teeth				
Ca	0.74 (0.54–1.01)	0.06	0.91 (0.63–1.32)	0.63
P	0.95 (0.72–1.24)	0.68	0.98 (0.72–1.33)	0.90
Mg	0.92 (0.73–1.16)	0.49	0.99 (0.76–1.30)	0.94
Ca/P ratio	0.49 (0.30–0.78)	0.003	0.52 (0.30–0.90)	0.02
DMFT index of permanent teeth				
Ca	0.76 (0.58–0.99)	0.04	0.85 (0.63–1.16)	0.30
P	0.94 (0.75–1.19)	0.62	0.87 (0.67–1.13)	0.30
Mg	1.08 (0.89–1.31)	0.42	1.23 (0.98–1.54)	0.07
Ca/P ratio	0.55 (0.37–0.82)	0.004	0.59 (0.37–0.93)	0.02

BMI = body mass index; Ca = calcium; CI = confidence interval; deft = d (decayed), e (indicated for extraction), f (filled) of primary teeth; DMFT = D (decayed), M (missing due to caries), F (filled) of permanent teeth; Mg = magnesium; P = phosphorus. DRIs = the Taiwanese Dietary Reference Intake values [10].

[a] Multiple logistic regressions adjusted by sex, age, birth order, parental education level, BMI, sweet intake, tooth brushing habits, and fluoride exposure.

changes in Ca/P ratio could also be due to other cariogenic or anticariogenic foods or substances. Apparently, based on the report of NAHSIT Children 2001–2002 [10], increasing the consumption of dark-green/yellow vegetables may increase the Ca/P ratio. It is still of interest for future studies to look into food items and sources related to the Ca/P ratio.

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